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Dogs of Nomadic Pastoralists in Northern Kenya Are Reservoirs of Plasmid-Mediated Cephalosporin- and Quinolone-Resistant *Escherichia coli*, Including Pandemic Clone B2-O25-ST131

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Resistance in *Escherichia coli* isolates colonizing gastrointestinal tracts of dogs, cats, and their owners in Northern Kenya was investigated with an emphasis on extended-spectrum beta-lactamases (ESBLs). Totals of 47 (22%, $n = 216$), 2 (4%, $n = 50$), and 4 (17%, $n = 23$) CTX-M-15-producing *E. coli* isolates were obtained from dogs, cats, and humans, respectively. CTX-M-15-producing *E. coli* isolates with identical PFGE profiles were detected in animals and humans living in the same area.

Transfer of resistant bacteria between humans and their dogs has been documented in various studies from the Western world (12). There have been no such data, however, from developing African countries. Dogs have been repeatedly recognized as comprising reservoirs or sentinels for zoonotic and livestock-infecting pathogens and, as such, are useful for epidemiological monitoring in rural parts of Africa (5). This study investigated the prevalence and molecular epidemiology of extended-spectrum beta-lactamase (ESBL)-producing *Escherichia coli* strains in Northern Kenya, an area inhabited mainly by nomadic herders of the Samburu, Turkana, El Molo, Rendille, and Gabra tribes. Although certain allopathic antimicrobials (mainly oxytetracycline) are occasionally used to treat livestock, veterinary medicine for small animals is virtually absent in the area (2). As direct selective antibiotic pressure is minimized in these animals, domestic carnivores can serve as sentinels of environmental contamination.

Rectal swabs of 216 dogs, 50 cats, and 23 humans were collected into Amies medium in September and October 2009 in nine settlements in Marsabit and Samburu Districts of Northern Kenya. Swabs were plated in parallel on plain MacConkey agar (MCA) (Oxoid, United Kingdom) and MCA with cefotaxime (2 mg/liter). One lactose-fermenting colony was isolated from each plate and tested for susceptibility to 12 antimicrobial agents using the disc diffusion method (8). Colonies obtained on cefotaxime-supplemented MCA were identified by using the API test (bioMérieux, France) and examined by the double-disc synergy test (8). The ESBL-positive *E. coli* isolates were tested by PCR and sequencing for (i) ESBL-coding genes bla_{TEM}, $bla_{\mathrm{CTX\text{-}M}}$, bla_{SHV}, and bla_{OXA}, (ii) plasmid-mediated quinolone-resistance genes *aac(6′)-Ib-cr*, *qepA*, *qnrA*, *qnrB*, *qnrC*, *qnrD*, and *qnrS*, and (iii) additional antibiotic resistance genes and integrons as described previously (14). All other methods used were described elsewhere (10). In brief, ESBL-producing *E. coli* were typed and clustered by pulse-field gel-electrophoresis (PFGE) (4) and their phylogenetic groups were identified by multiplex PCR assay (6). In isolates belonging to phylogenetic group B2, allele-specific PCR was performed to identify the O25-ST131 clone (7). Multilocus sequence typing (MLST) determination was carried out in isolates positive by allele-specific PCR and analyzed at http:/mlst.ucc.ie/mlst/dbs/Ecoli (17). The insertion sequence IS*EcpI* in the upstream region of *bla* genes was tested (11). Chemical transformation of plasmids and conjugation experiments to *E. coli* MT102RN and *Salmonella enterica* serovar Typhimurium SL5325 were performed (15). Plasmids were analyzed by S1-PFGE (1), replicon typing (3), and restriction fragment length polymorphism profiles obtained by EcoRV digestion.

A total of 267 lactose-fermenting isolates were obtained by cultivating the swabs on MCA without antibiotics. The resistance patterns of these isolates ranged from 1 (ampicillin, gentamicin, tetracycline, streptomycin, or nalidixic acid) to all 12 of the antibiotics tested. Multiresistance to ampicillin, streptomycin, sulfonamides, tetracycline, and trimethoprim-sulfamethoxazole was the most frequent phenotype.

Totals of 47 (22%, $n = 216$), 2 (4%, $n = 50$), and 4 (17%, $n = 23$) ESBL-positive *E. coli* isolates were obtained from dogs, cats, and humans, respectively, by cultivation on cefotaxime-supplemented MCA. Resistance to amoxicillin-clavulanic acid was found in 15% and resistance to ceftazidime in 11% of the ESBL-producing isolates. Apart from resistance to beta-lactam antibiotics, these isolates showed resistance to tetracycline (100% of the ESBL-producing isolates), trimethoprim-sulfamethoxazole (100%), sulfonamides (98%), nalidixic acid (96%), ciprofloxacin (94%), gentamicin (87%), chloramphenicol (43%), and streptomycin (34%). Comparison of PFGE banding patterns clustered the ESBL-producing isolates into five groups of closely related isolates (>85% similarity) designated clusters K, L, M, N, and O (Fig. 1). Overall, 36 (70%) of the ESBL-producing isolates belonged to either cluster K or L. Isolates with identical PFGE profiles were detected from 8 dogs and 2 humans, and closely related (>95% similarity) isolates were found in 1 human and 1 cat sampled in the same town. Three dogs were found to harbor isolates of

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TABLE 1 Strain and plasmid characteristics of ESBL-producing *E. coli* isolates *from* dogs, cats, and humans[a]

	Characteristics of:								
	Strain					Plasmid[c]			
Locality	Source and identifier	PFGE cluster	Phylogroup, sequence type	Resistance phenotype	Resistance genes in addition to β-lactamases[b]	Conjugation or transformation	Size (kb), Inc group	Genes cotransferred with $bla_{CTX-M-15}$ and $bla_{OXA1-like}$	EcoRV restriction pattern
Gas	Dog 231	L	A	Am St Su Te Sx Cm Cf Na Gn Cp	*sul2*, *catA1*, *tet*(B), *dhfr12*, *dhfr1*, *strA*, *aac(6′)-lb-cr*				
	Dog 233	L	A	Am St Su Te Sx Cf Cz	*int1*, *sul2*, *tet*(B), *dhfr1*, *strA*, *aac(6′)-lb-cr*	Conj *E. coli*	160, FIA/FIB	*int1*, *aac(6′)-lb-cr*	4
	Dog 235	K	A	Am St Su Te Sx Cf Na Cz Gn Ac Cp	*int1*, *sul2*, *catA1*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 232	X	A	Am St Su Te Sx Cf Cz	*sul2*, *tet*(B), *aadA2*, *strA*, *aac(6′)-lb-cr*				
Arapal and Gatab[d]	Dog 166	K	A	Am Su Te Sx Cm Cf Na Cz Gn Cp	*int1*, *sul2*, *catA1*, *tet*(B), *strA*, *aac(6′)-lb-cr*	Trans, conj *E. coli*	160, FIA/FIB	*int1*, *sul2*, *tet*(B), *catA1*, *aac(6′)-lb-cr*	3
	Dog 20	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *catA1*, *tet*(B), *strA*, *aac(6′)-lb-cr*				
	Dog 36	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 49	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *catA1*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 89	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 38	M	A	Am St Su Te Sx Cm Cf Na Gn Ac Cp	*sul2*, *catA1*, *cml*, *tet*(B), *aac(6′)-lb-cr*				
	cat 346	X	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *catA1*, *tet*(B), *aadA2*, *aac(6′)-lb-cr*	Trans, conj *E. coli*	160, FIA/FIB	*int1*, *sul2*, *tet*(B), *catA1*, *aac(6′)-lb-cr*	3
South Horr	Dog 168	N	B2, ST131	Am Su Te Sx Cf Na Cp	*int1*, *sul2*, *tet*(A), *aac(6′)-lb-cr*	Conj *E. coli*			
	Dog 198	N	B2, ST131	Am Su Te Sx Cf Na Cp	*int1*, *sul2*, *tet*(A), *aadA2*, *aac(6′)-lb-cr*				
	Dog 209	N	B2, ST131	Am St Su Te Sx Cf Na Ac Cp	*int1*, *sul2*, *tet*(A), *strA*, *aac(6′)-lb-cr*				
	Dog 205	X	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *tet*(B), *aac(6′)-lb-cr*				
Loyiangalani	Dog 62	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *catA1*, *tet*(B), *aac(6′)-lb-cr*	Trans	160, FIA/FIB	*int1*, *sul2*, *tet*(B), *catA1*, *aac(6′)-lb-cr*	3
	Dog 110	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *catA1*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 119	K	A	Am Su Te Sx Cm Cf Na Gn Ac Cp	*int1*, *catA1*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 128	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 130	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*catA1*, *sul2*, *tet*(B), *strA*, *aac(6′)-lb-cr*	Trans, conj *E. coli*	110, FIB	*sul2*, *tet*(B), *catA1*, *aac(6′)-lb-cr*	1
	Dog 131	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *catA1*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 135	K	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *catA1*, *tet*(B), *strA*, *aac(6′)-lb-cr*				
	Dog 154	K	A	Am St Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *catA1*, *tet*(B), *strA*, *aac(6′)-lb-cr*				
	Dog 239	K	A	Am St Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *catA1*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 242	K	A	Am Su Te Sx Cm Cf Na Gn Ac Cp	*int1*, *sul2*, *tet*(B), *dhfr12*, *aac(6′)-lb-cr*	Conj *E. coli*			
	Dog 7	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aac(6′)-lb-cr*	Conj *E. coli*	160, FIA/FIB	*sul2*, *int1*, *aac(6′)-lb-cr*	5
	Dog 41	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *strA*, *tet*(B), *dhfr1*, *aac(6′)-lb-cr*				
	Dog 42	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *dhfr1*, *aac(6′)-lb-cr*				
	Dog 43	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *dhfr1*, *aac(6′)-lb-cr*				
	Dog 115	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 136	L	A	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *strA*				
	Dog 137	L	A	Am St Su Te Sx Cf Na	*int1*, *sul2*, *tet*(B), *aadA1*, *strA*, *aac(6′)-lb-cr*				
	Dog 140	L	A	Am Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aadA1*	Conj *E. coli*			
	Dog 146	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aac(6′)-lb-cr*				
	Dog 150	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *strA*, *aac(6′)-lb-cr*				

Dog 156	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aadA1*, *strA*, *aac(6′)-lb-cr*				
Dog 229	L	A	Am St Su Te Sx Cf Na Cz Gn Cp	*int1*, *sul2*, *tet*(B), *dhfr1*, *strA*, *aac(6′)-lb-cr*	Conj *E. coli*	150, FIA/FIB	*int1*, *aac(6′)-lb-cr*, *catA1*	2
Dog 237	L	A	Am Su Te Sx Cf Na Cz Gn Ac Cp	*int1*, *sul2*, *tet*(B), *aac(6′)-lb-cr*	Conj *S.* Typhimurium			
Dog 238	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aac(6′)-lb-cr*				
Dog 240	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aac(6′)-lb-cr*	Conj *E. coli*	150, FIB	*int1*, *tet*(B), *aac(6′)-lb-cr*	2
Human 1	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aac(6′)-lb-cr*				
Human 18	L	A	Am Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *dhfr12*, *aac(6′)-lb-cr*	Conj *E. coli*	150, FIB	*int1*, *tet*(B), *aac(6′)-lb-cr*	2
Human 23	L	A	Am St Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *strA*, *aac(6′)-lb-cr*	Conj *E. coli*	150, FIB	*int1*, *tet*(B), *aac(6′)-lb-cr*	2
Dog 116	M	A	Am St Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *catA1*, *tet*(B), *aadA1*, *strA*, *aac(6′)-lb-cr*	Conj *E. coli*, *S.* Typhimurium	110, FIB	*sul2*, *tet*(B), *catA1*, *aac(6′)-lb-cr*	1
Dog 124	M	A	Am Su Te Sx Cf Na Cz Gn Cp	*int1*, *sul2*, *tet*(B), *strA*, *aac(6′)-lb-cr*				
Dog 126	M	A	Am St Su Te Sx Cm Cf Na Gn Cp	*sul2*, *catA1*, *tet*(B), *strA*, *aac(6′)-lb-cr*	Trans, conj *E. coli*	110, FIB	*sul2*, *tet*(B), *catA1*, *aac(6′)-lb-cr*	1
Dog 138	M	A	Am St Su Te Sx Cm Cf Na Gn Ac Cp	*catA1*, *sul2*, *tet*(B), *strA*, *aac(6′)-lb-cr*				
Dog 114	O	B1	Am St Su Te Sx Cp	*int1*, *tet*(B), *aac(6′)-lb-cr*				
cat 344	O	B1	Am St Su Te Sx Cf Na Cz Gn Ac Cp	*int1*, *tet*(A), *tetD*, *strA*, *aac(6′)-lb-cr*				
Human 12	O	B1	Am St Su Te Sx Cm Cf Na Gn Cp	*int1*, *catA1*, *tet*(B), *aadA1*, *strA*, *aac(6′)-lb-cr*	Conj *E. coli*			
Dog 141	X	B1	Am Su Te Sx Cm Cf Na Gn Cp	*int1*, *sul2*, *catA1*, *cml*, *tet*(A), *strA*, *aac(6′)-lb-cr*	Conj *E. coli*	90, I1	*sul2*	
Dog 153	X	B1	Am Su Te Sx Cf Na Gn Cp	*sul1*, *tet*(B), *aac(6′)-lb-cr*				
Human 15	X	A	Am St Su Te Sx Cf Na Gn Cp	*int1*, *sul2*, *tet*(B), *aadA1*, *dhfr12*, *strA*, *aac(6′)-lb-cr*	Conj *E. coli*	150, FIB	*int1*, *tet*(B), *aac(6′)-lb-cr*	2

[a] Am, ampicilin; St, streptomycin; Su, sulfonamide compounds; Te, tetracycline; Sx, trimethoprim-sulfamethoxazole; Cm, chloramphenicol; Cf, cephalotin; Na, nalidixic acid; Gn, gentamicin; Cp, ciprofloxacin; Cz, ceftazidime; Ac, amoxycilin-clavulanate.

[b] All strains harbored the $bla_{\text{CTX-M-15}}$ gene and the $bla_{\text{OXA-1-like}}$ gene. *int1* is integron 1 with *dfrA17-aadA5* gene cassettes.

[c] Plasmid characteristics are stated for those transformants/transconjugants which harbored one plasmid. Conj, conjugation; Trans, transformation.

[d] Arapal and Gatab are close to each other, so the samples were grouped together.

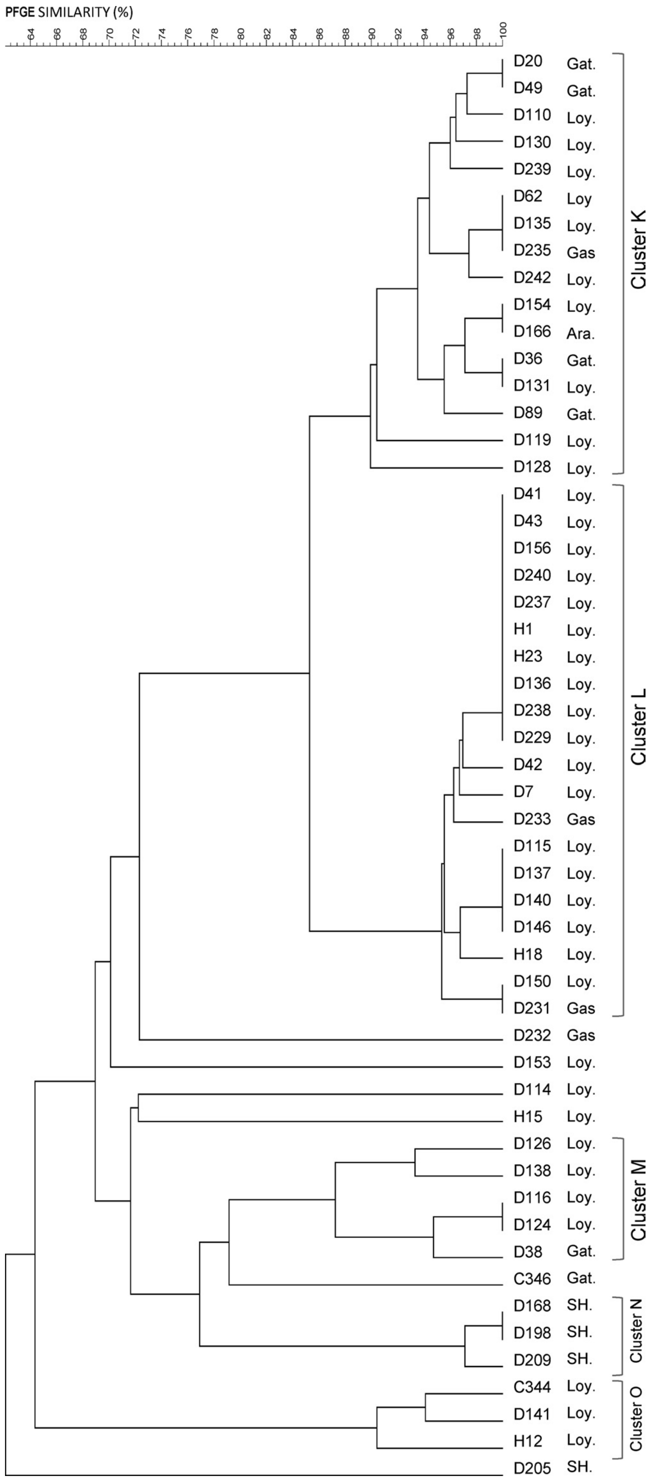

FIG 1 Dendrogram of CTX-M-producing *E. coli* isolates' PFGE profiles, generated by cluster analysis of the Dice similarity indices in the BioNumerics fingerprinting software (optimization 1%, band matching tolerance 1%, tolerance change 1%). Gat, Gatab; Loy, Loyiangalani; Gas, Gas; Ara, Arapal; SH, South Horr. D, dog; H, human; C, cat.

the B2-O25-ST131 lineage. The genes $bla_{\text{CTX-M-15}}$ and $bla_{\text{OXA-1-like}}$ were found in all ESBL-producing *E. coli* isolates; all of these isolates also tested positive for the gene *aac-(6′)-Ib-cr*, but none of them had *qnr* genes. The upstream region of $bla_{\text{CTX-M-15}}$ in all ST131 isolates contained the transposase gene of the IS*26* sequence. All other isolates had the IS*Ecp1* insertion sequence upstream from the $bla_{\text{CTX-M}}$ gene, but PCR for IS*26* was negative. Five distinct types of plasmids were detected: (i) a 90-kb plasmid of incompatibility group I1 (IncI1), (ii) a 110-kb plasmid of group IncFIB, (iii) a 150-kb plasmid of group IncFIB, (iv) a 150-kb plasmid of group IncFIA and IncFIB (IncFIA/FIB), and (v) a 160-kb plasmid of group IncFIA/FIB (Table 1). All these plasmids carried the $bla_{\text{CTX-M-15}}$ and *aac(6′)-Ib-cr* genes together with various combinations of other resistance genes, *dfrA17-aadA5*, *tet*(B), or *catA1*. Plasmids of the 160-kb, IncFIA/FIB, and identical EcoRV profile carrying *int1*, *tet*(B), *catA1*, and *aac(6′)-Ib-cr* were found in PFGE-unrelated isolates from dogs and a cat sampled in different villages. The EcoRV restriction fragment length polymorphism (RFLP) patterns of all IncF plasmids shared more than 85% similarity (difference in up to five bands).

The findings of this study corroborate the recognized worldwide spread of CTX-M-15-producing isolates, including the O25-ST131 clone, even in very remote areas where extended spectrum cephalosporins are used very rarely (9, 13, 16). The $bla_{\text{CTX-M-15}}$ gene was found on large conjugative plasmids together with genes encoding resistance to other groups of antibiotics. Such a genetic constellation, along with the warm climate and lack of sanitary facilities in rural Africa, probably facilitates the spread of multiresistant bacteria and their transfer between humans and domestic carnivores. The relatively lower resistance rates in isolates from cats compared to the resistance rates in isolates from dogs could be explained by the different foraging behaviors of the two species. While cats may prefer hunting in the bush, dogs are probably more reliant on household leftovers and more prone to coprophagy. Such a scavenger strategy renders dogs good sentinels of environmental contamination and suitable for monitoring of local resistance patterns. Further studies are needed to specify the role of livestock, the mainstay of nomadic pastoralist communities, as well as omnipresent domestic rodents, in the epidemiology of antibiotic-resistant bacteria.

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REFERENCES

1. **Barton BM, Harding GP, Zuccarelli AJ.** 1995. A general method for detecting and sizing large plasmids. Anal. Biochem. **226**:235–240.
2. **Bett B, Jost Cand Mariner J.** 2008. Participatory investigation of important animal health problems amongst the Turkana pastoralists: relative incidence, impact on livelihoods and suggested interventions. Discussion Paper No. 15. Targeting and Innovation. International Livestock Research Institute, Nairobi, Kenya.
3. **Caratolli A, et al.** 2005. Identification of plasmids by PCR-based replicon typing. J. Microbiol. Methods **63**:219–228.
4. **CDC.** 2004. Standardized molecular subtyping of foodborne bacterial pathogens by pulse-field gel electrophoresis. Centers for Disease Control, Atlanta, GA.

5. **Cleaveland S, Meslin FX, Breiman R.** 2006. Dogs can play useful role as sentinel hosts for disease. Nature **440**:605.
6. **Clermont O, Bonacorsi S, Bingen E.** 2000. Rapid and simple determination of *Escherichia coli* phylogenetic group. Appl. Environ. Microbiol. **66**: 4555–4558.
7. **Clermont O, et al.** 2009. Rapid detection of the O25-ST131 clone of *Escherichia coli* encompassing the CTX-M-15-producing strains. J. Antimicrob. Chemother. **64**:274–277.
8. **CLSI.** 2008. Performance standards for antimicrobial susceptibility testing, 18th informational supplement. CLSI document M100-S18. CLSI, Wayne, PA.
9. **Coque TM, et al.** 2008. Dissemination of clonally related *Escherichia coli* strains expressing extended-spectrum beta-lactamase CTX-M-15. Emerg. Infect. Dis. **14**:195–200.
10. **Dolejska, M., et al.** CTX-M-15-producing *Escherichia coli* clone B2-O25b-ST131 and *Klebsiella* spp. in municipal wastewater treatment plant effluents. J. Antimicrob. Chemother. **66**:2784–2790.
11. **Eckert C, Gautier V, Arlet G.** 2006. DNA sequence analysis of the genetic environment of various $bla_{\text{CTX-M}}$ genes. J. Antimicrob. Chemother. **57**: 14–23.
12. **Guardabassi L, Schwarz S, Lloyd DH.** 2004. Pet animals as reservoirs of antimicrobial-resistant bacteria. J. Antimicrob. Chemother. **54**:321–332.
13. **Hernandez J, et al.** 2010. Globally disseminated human pathogenic *Escherichia coli* of O25-ST131 clone, harboring $bla_{\text{CTX-M-15}}$, found in glaucous-winged gull at remote Commander Islands, Russia. Environ. Microbiol. Rep. **2**:329–332.
14. **Literak I, et al.** 2010. Antimicrobial-resistant faecal *Escherichia coli* in wild mammals in central Europe: multiresistant *Escherichia coli* producing extended-spectrum beta-lactamases in wild boars. J. Appl. Microbiol. **108**: 1702–1711.
15. **Olesen I, Hasman H, Aarestrup FM.** 2004. Prevalence of beta-lactamases among ampicillin-resistant *Escherichia coli* and *Salmonella* isolated from food animals in Denmark. Microb. Drug Resist. **10**:334–340.
16. **Pallecchi L, et al.** Rapid dissemination and diversity of CTX-M extended-spectrum beta-lactamase genes in commensal *Escherichia coli* isolates from healthy children from low-resource setting in Latin America. Antimicrob. Agents Chemother. **51**:2720–2725.
17. **Wirth T, et al.** 2006. Sex and virulence in *Escherichia coli*: an evolutionary perspective. Mol. Microbiol. **60**:1136–1151.